REMARKS

Applicant respectfully requests that the Examiner enter the foregoing amendments and consider the following remarks.

Claims 14-17 are requested to be cancelled.

Claims 18-21 are being added.

This amendment adds, changes and/or deletes claims in this application. A detailed listing of all claims that are, or were, in the application, irrespective of whether the claim(s) remain under examination in the application, are presented, with an appropriate defined status identifier. The cancellation of claims herein is intended only to remove non-elected claims, and does not constitute an admission that any of the subject matter of the canceled claims is unpatentable. Applicant reserves the right to prosecute claims to such subject matter in any appropriate application.

After amending the claims as set forth above, claims 1-13 and 18-21 are now pending in this application.

Applicant appreciates the Examiner's indication that claims 5-8 and 10-13 are allowable. Nonetheless, as explained below, Applicant respectfully submits that additional claims are also properly allowable, and requests reconsideration and withdrawal of the outstanding rejections.

Applicant respectfully requests that the Examiner fully consider the remarks below in connection with the outstanding rejections of the claims as allegedly being obvious. As discussed below, the Examiner's comments in the Advisory Action mailed June 30, 2003 suggests that the Examiner did not fully consider the significance of the points and prior art discussed by Applicant, which may have led the Examiner to maintain the rejections.

Sequence Listing

As required by the Examiner, a replacement Sequence Listing that includes the sequence of Fig. 1 was submitted in this application with the prior, non-entered amendment. Applicant requests that the previously submitted Sequence Listing be entered in the specification.

Rejections under 35 USC § 103

The Examiner rejected claims 3 and 9 under 35 USC § 103(a) as allegedly being anticipated by Evangelista et al. (Anal. Biochem. (1996) 235:89-97), rejected claims 1-3 and 9 under 35 USC § 103(a) over Evangelista in view of Tabor et al. (U.S. Patent 5,614,365), and rejected claims 3, 4, and 9 under 35 USC § 103(a) as allegedly being unpatentable over Evangelista in view of Haralambidis et al. (NAR (1987) 15(12):4857-4876).

The Examiner asserted that Evangelista teaches a compound of formula (I) (A-B-C) with a cyanine dye which meets the structural requirements of A, a linker of more than 10 atoms in length as required by B, and attached to the 5-position of dUTP, which has a triphosphate attached. In connection with claims 3, and 9, the Examiner further asserted that it would have been *prima facie* obvious to modify the labeled deoxynucleotide of Evangelista into a dideoxynucleotide since Evangelista et al. note that dNTPs and ddNTPs are employed in DNA sequencing techniques. In connection with claims 1-3 and 9, the Examiner asserted that it would have been "*prima facie* obvious to combine the fluorescently labeled ddNTPs which are made obvious by Evangelista into a kit with the modified thermostable polymerases of Tabor." In connection with claims 3, 4, and 9, the Examiner asserted that it would have been *prima facie* obvious to use the linker described in Haralambidis et al in the present claimed dye terminator molecules, further asserting that an ordinary practitioner would have been motivated to utilize the long linker art of Harambidis in the synthesis of the cyanine dye of Evangelista for the expressly stated benefits of mild conditions, high yield and efficient hybridization. Applicant respectfully traverses these rejections.

Preliminarily, while both dNTPs and ddNTPs can be employed in DNA sequencing, labelled dNTPs and labelled ddNTPs are used very differently, the roles of these two different

molecules in sequencing are quite different, and their suitability for use with various DNA polymerases can differ dramatically. As explained in the Background of the specification and in Tabor, U.S. Patent No. 5,614,365 (col. 3, lines 9-15), most DNA polymerases discriminate to a substantial degree against the incorporation of ddNTPs relative to dNTPs, and the degree of discrimination varies with the sequence context, often leading to high variability in sequencing gel band intensities. As pointed out in the present specification, such discrimination and resulting band intensity variability also exists in use of dye-labelled ddNTPs, with the effects exacerbated by the presence of the dye moiety. The result of the differences in use and behavior between dNTPs and ddNTPs means that uses described for particular labeled dNTPs do not suggest that the ddNTPs with the same labels should be made and used in sequencing as dye terminators.

This lack of suggestion specifically applies to the Evangelista et al. reference cited by the Examiner. As previously discussed, Evangelista et al. is the central reference for each of the Examiner's rejections; if the alleged *prima facie* case of obviousness of the present dye terminator molecules is not made based on Evangelista, then each of the combinations of references cited by the Examiner will also fail to make a *prima facie* case of obviousness. Applicant respectfully submits that the alleged *prima facie* case of obviousness is not made, because Evangelista et al. does not suggest making the claimed dideoxy dye terminator molecules, nor does it (alone or in combination with either of the additional cited references) suggest providing a kit containing 4 different dideoxy dye terminators that each include a linker of at least 10 atoms between the dye and the nucleotide base, and a thermostable DNA polymerase.

Prior to the present invention, no one had recognized that the length of the linker in dye-labeled dideoxynucleotide triphosphates (ddNTPs) was an important and consistent variable in providing more uniform band intensities and resolving dye-induced compression artifacts in DNA sequencing. (See, Specification, p.5, lines 7-9.) As described in the Background section of the present Specification, certain dye-labeled ddNTP terminators had been produced, but dye-

induced compression-like artifacts were observed in many cases with such terminators, even when linkers were used and even when using Taq DNA polymerase and modified T7 DNA polymerase. Typically, attempts to resolve such artifacts involved the use of modified NTPs. (See, Specification, p.2, line 7 to p.4.)

Despite such reports on the use of dye labeled ddNTPs and the failure of the various investigators to recognize the importance of the effect of linker length on band uniformity, the present inventors surprisingly discovered "that there is a strong correlation between the length of the link between the dye molecules and the nucleotide, but little correlation between the type of dye (or other parameters) and band uniformity", and that extended linkers with 10-25 atoms provide "significantly improved uniformity compared with dye terminators with linkers less than 10 atoms." (Specification, p.5, lines 17-27 (emphasis added).) In keeping with this discovery, the present inventors found that the present novel molecules can be used for sequencing to provide significantly improved band uniformity in sequencing gels, without the need to use nucleotide analogs.

In contrast to the use of dye-labeled ddNTPs as dye terminators as described in the present invention, Evangelista et al. describes the use of labeled dNPTs in filling recessed 3'-ends using Klenow fragment. Despite the failure of Evangelista et al. to provide any indication that dideoxy dye terminators as presently claimed should be made and used in DNA sequencing, in connection with claims 3 and 9, the Examiner asserted that it would have been *prima facie* obvious to modify the labeled deoxynucleotide of Evangelista into a dideoxynucleotide. In order for such a *prima facie* case to be made, it is necessary that there be a suggestion from the prior art to make the modifications leading to the claimed invention.

The requirement for such a suggestion is well-established by Federal Circuit opinions. Whether involving a single reference or a combination of references, in order to make an invention obvious, there must be a suggestion or motivation from the prior art to make the modifications to provide the claimed invention, and such suggestion or motivation must be supported by evidence. *In re Sang-Su Lee*, 61 USPQ2d 1430 (Fed. Cir. 2002) (citing *In re*

Kotsab, 55 USPQ2d 1313, 1317 (Fed. Cir. 2000) ("particular findings must be made as to the reason the skilled artisan, with no knowledge of the claimed invention would have selected those components for combination in the manner claimed")). In addition, the basis and reasons for concluding that an invention is obvious must be clearly explained. Thus, the Federal Circuit has stated that the PTO "must explain the reasons one of ordinary skill in the art would have been motivated to select the references and to combine them to render the claimed invention obvious. *Id.* (citing *In re Rouffet*, 47 USPQ 2d 1453, 1459 (Fed. Cir. 1998)). In short, "the showing must be clear and particular." *In re Dembiczak*, 50 USPQ2d 1614 (Fed. Cir. 1999). Applicant respectfully submits that neither the requisite suggestion, nor a clear explanation of the Examiner's basis for finding such a suggestion in present in this case.

It appears that the Examiner's assertion that it would have been *prima facie* obvious to modify the labeled deoxynucleotide of Evangelista into a dideoxynucleotide is not supported. This assertion ignores the limitations in the Evangelista et al. description, and the context for sequencing, *e.g.*, as described in the Background of the present specification and in Tabor, US Patent 5,614,365. There is simply no suggestion from Evangelista to make the present claimed dye terminators and use them in sequencing, and such a suggestion is essential support a *prima facie* case of obviousness.

In response to prior arguments, the Examiner provided discussion allegedly justifying maintenance of the rejections. The Examiner asserted that "Evangelista clearly envisions the use of the labeled nucleotides in automated sequencing type asssay(see page 94, column 1). Such assays routinely use labeled ddNTPs for labeling. So when Evangelista teaches the label and suggests the use of the label in sequencing type reactions, this is a direct suggestion to make labeled ddNTPs." Applicant respectfully submits that this assertion misconstrues the Evangelista disclosure.

Evangelista specifically limits the discussion to the use of fluor-labeled dNTPs "in automated sequencing with internal labeling using fluorescein-labeled dNTP and in nick translation and random priming using cyanine-labeled dNTPs." (page 94, paragraph bridging

cols. 1 & 2) Neither of these processes relates to the use of cyanine dye-labeled ddNTPs. Therefore, this citation in Evangelista provides no more suggestion concerning the present novel dye terminators than it does about novel DNA polymerases that may be used in automated sequencing. There is simply nothing in Evangelista that suggests jumping the gap from dNTPs to ddNTPs in view of the differences in reaction in enzymatic polymerization and in sequencing gel results.

As explained in the Background of the present specification (and pointed out above), the present invention addressed a particular issue connected with the use of dye-labeled ddNTPs, that of lack of band uniformity in sequencing gels. The solution to this issue suggested by the prior art was that of using nucleotide analogs. In addition to being directed to the use of dNTPs, Evangelista is simply not concerned with gel band problems associated with use of dideoxy dye terminators or with a sequencing method that uses dideoxy dye terminators. Thus, Evangelista cannot suggest solutions to a problem that arises when using fluor-labeled ddNTPs in sequencing where neither the process not the problem is discussed in the reference.

The lack of such suggestion from Evangelista as well as other cited references is, in fact, further evidenced by the failure of others to develop the claimed dideoxy dye terminators. As pointed out above, various other dye-labeled ddNTPs had been prepared, even prior to publication of the Evangelista article (see Background of present Specification). As specific examples, Prober, *Science* 238:336-342 was published in 1987; Lee, *Nucl. Acids Res.*, 20:2471-2483 was published in 1992. In addition, Evangelista pointed out that some of the labeled dNTPs used in the tests were commercially available. Despite the passage of 11 years from the Prober publication, 6 years from the Lee publication, and 4 years from the Evangelista publication to the filing of the parent of the present application, the present inventors were the first to make and use the present dye terminator molecules, being the first to discover the advantageous effects of using extended linkers for dye-labeled ddNTPs. The passage of such periods of time is further demonstration of the lack of suggestion of the present invention.

In accordance with the discussion above, the disclosures in the references described in the Background of the present Specification, and the disclosure in Evangelista, it is apparent that the solution to lack of uniformity in sequencing gel bands involving dye labeled dideoxy terminators was directed to the use of various analogs and that Evangelista did not address or even acknowledge this issue. Thus, Evangelista provides no suggestion or motivation to provide the present molecules and kits. Since there is no suggestion from the art cited by the Examiner to make the claimed dideoxy dye terminators, or to provide such molecules in sequencing kits, the present claims cannot be obvious under controlling Federal Circuit law.

Despite the discussion above (which was in large part submitted in the Amendment filed May 21, 2003) and the previous explanation provided by Applicant, in the Advisory Action mailed June 30, 2003, the Examiner asserted that the statement in Evangelista et al. that " 'Fluro-labeled deoxynucleotide triphosphates (dNTPs) or dideoxynucleoside triphosphates (ddNTPs) are employed in nonradioactive DNA sequencing techniques such as those developed by Prober et al (ref omitted) and Ansorge et al (ref omittedP as well as for incorporation into hybridization probes (ref omitted). Fluorescent ddNTPs have also been used as terminal deoxynucleotidyl transferase substrates to label single (ref omitted) and double stranded DNA (ref omitted)' " constituted an "express suggestion" to form fluorescent ddNTPs for sequencing. Applicant respectfully submits that the Examiner's assertion is unreasonable and is simply incorrect. Applicant is unable to discern how a statement introducing some background on the use of fluor-labeled dNTPs and ddNTPs is somehow metamorphosed into an "express suggestion". It appears that the Examiner is failing to consider the Evangelista reference in its entirety including its limitations, and is similarly failing to consider the additional art, e.g., as discussed above, that indicates that the Evangelista would not, in fact, suggest the present invention to one of ordinary skill in the art.

Thus, Applicant respectfully requests that the Examiner properly reconsider the rejections in view of the entirety of evidence.

Applicant submits that the present application is now in condition for allowance. Favorable reconsideration of the application as amended is respectfully requested.

The Examiner is invited to contact the undersigned by telephone if it is felt that a telephone interview would advance the prosecution of the present application.

The Commissioner is hereby authorized to charge any additional fees which may be required regarding this application under 37 C.F.R. §§ 1.16-1.17, or credit any overpayment, to Deposit Account No. 50-0872. Should no proper payment be enclosed herewith, as by a check being in the wrong amount, unsigned, post-dated, otherwise improper or informal or even entirely missing, the Commissioner is authorized to charge the unpaid amount to Deposit Account No. 50-0872. If any extensions of time are needed for timely acceptance of papers submitted herewith, applicant hereby petitions for such extension under 37 C.F.R. §1.136 and authorizes payment of any such extensions fees to Deposit Account No. 50-0872.

Respectfully submitted,

Date 21 June 2003

By 

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